REMARKS

The Invention

The present invention provides new methods for treating disease by administering a protease that has been mutated to cleave a target protein involved in the disease pathology, as well as methods for generating and screening for protease muteins useful in the method. In one important embodiment of the method, the specificity of the protease is altered such that it cleaves a non-cognate sequence in the target protein. As a result of a restriction requirement, prosecution of the present application is focused on claims for identifying protease muteins useful in the method.

Procedural Status

Claims 1-16 and 45-58 have been rejected as follows:

(1) under 35 U.S.C. § 112, first paragraph and second paragraph, for failing to comply with the written restriction requirement, with the enablement requirement, as indefinite,

(2) under 35 U.S.C. § 103, as obvious over certain references cited by the Examiner. To speed allowance of claims drawn to important subject matter, the Applicants have amended the claims without prejudice or disclaimer, added new claims to address the Examiner's concerns as stated in the rejection, and so request favorable reconsideration of the application and withdrawal of all rejections. In particular, the Applicants have amended certain, and/or added, claims that include a recitation of the types of target proteins that can be targeted by the mutated and altered muteins identified in the method, that include a recitation of the types of proteases that can be mutated, and that include both recitations.

The Examiner also seemed concerned that the claims did not recite the locations in a target protease that would have to be mutated to create a protease of the desired increased cleavage activity or altered specificity. As the Applicants explain below, one of the key benefits of the present inventive method is that one does not have to know where to mutate a protease to achieve the desired activity or specificity profile to identify a mutant protease having the desired features. Accordingly, the Applicants believe that the present claims, which allow one to screen a wide variety of mutein proteases to identify proteases that may have important therapeutic activity completely different from the activity of the wild-type protease from which they were

derived, are drawn to an invention our patent laws were designed to award with patent protection.

Claim Amendments

Claims 1-16 and 45-58 were pending prior to the present amendment. Claims 17-44 have been canceled without prejudice or disclaimer as drawn to a non-elected invention.

In this Amendment, Claims 8, 10, 49 and 55 have been canceled. Claims 1, 7, 9, 12, 13, 14, 15, 16, 45-47, 51, 53, 54, 57 and 58 have been amended. New claims 59-66 have been added. Support for the amended and new claims is given below.

Specifically, claims 1 and 53, as amended, focus on mammalian proteases (see p. 17, line 28 of the specification) that cleave target proteins involved with a pathology such that cleavage of the target at a substrate sequence recognized by the protease mutein provides a treatment for the pathology (see p. 9, lines 26-28, of the specification). These amendments have rendered redundant Claims 8, 10 and 55, which accordingly have been canceled and amendment of Claim 9 to depend from Claim 1, instead of Claim 8. Applicants have herein amended Claims 1 and 53 to specify that the mutein protease has an increased cleavage activity and/or an altered substrate specificity for cleaving the substrate sequence, relative to the wild-type scaffold sequence. See specification, p. 9, line 10-25; p.16, lines 6-7; p. 17, lines 8-10. Specificity and activity of a protease are discrete concepts (see, page 8, line 18, which states that mutations in the scaffold of an existing protease change the specificity and activity of the protease). Cleavage activity includes the catalysis of peptide bond hydrolysis of a given polypeptide. The specification discloses the measurement of cleavage activity at, *e.g.*, Example 4 on page 45, lines 1-12, wherein the cleavage of TNF and TNF receptors is determined. Substrate specificity pertains to the cleavage of a target polypeptide by a protease at a specific site within the protein.

The present invention relates in part to modifying a protease to generate a mutein that has an altered substrate specificity, meaning that the panoply of polypeptide sequences acted upon by, and/or the activity on each of the sequences in that panoply of, the mutein protease is changed relative to the wild-type protease. For example, muteins having altered substrate specificity relative to a wild-type protease are capable of acting on a polypeptide sequence not recognized by or only weakly recognized by the wild-type protease. However, if the mutein is still able to cleave the polypeptide sequence that is a target for the wild-type protease, the cleavage activity of the mutein enzyme may not be increased. The specification discloses the determination of wild-

type and mutein specificity activity at, *e.g.*, Example 11 on pages 51-54. For example, the I99A/N218A granzyme B mutein has altered specificity for the target polypeptide, caspase-3, relative to the wild-type protease. See Figures 3A-B.

Claim 1 has been amended to recite that the target protein is a cell surface molecule that transmits an extracellular signal for cell proliferation, a cytokine, a cytokine receptor, and a signaling protein that regulates apoptosis (see, e.g., p. 10, line 6, lines 22-24 and lines 29-30, p. 17, line 33 - p. 18, line 1).

Claim 53 has been amended to recite that the protease is selected from a specifically recited group of proteases. See, e.g., Table 3, p. 21.

Finally, to address the Examiner's concerns about the clarity of these claims, Claims 1 and 53 have been amended to recite that the wild-type scaffold sequence is the sequence of the "mammalian protease" from which the muteins differ by the recited number of N mutations.

As noted above, the amendments to Claims 1 and 53 are fully supported by the specification as filed.

Claim 7 (which depends from claim 1) has been amended to recite specific protease scaffold sequences -- the same group as recited in amended claim 53 (see, e.g., Table 3, p. 21).

Claims 9 and 49 have been amended to depend from Claim 1.

Claim 12 has been amended to recite specific target protein substrate sequences (see, e.g., Table 1, p. 12 -- VEGF; p. 50, Example 10, -- VEGF-R and p. 51, Example 11 -- caspase 3).

Claims 13-15 have been amended to recite that the specificity of the protease is increased by the recited amounts (see p. 17, lines 10, 12, and 14, respectively, in the specification).

Claim 16 has been amended additionally to recite altered specificity (as with the amendments to claims 1 and 53).

Claims 45-47 have been amended for clarity in light of the amendments to the claims from which they depend.

New independent Claim 59, and new Claims 60-62, which are dependent thereon, have been added. New Claim 59 contains the subject matter of amended Claim 1 and the additional recitation of human protease muteins and recites that the protease is selected from the specifically recited group of proteases (see p. 3, lines 10-16). New dependent Claim 60 recites that step (c) of Claim 59 is accomplished by identifying at least one protease mutein having altered substrate specificity for cleaving said substrate sequence, relative to the wild-type scaffold sequence.

New dependent Claim 61 recites a small Markush group of specific human protease muteins (supported throughout the specification). New Claim 62 recites specific target proteins (as in original claim 40 and p. 4, lines 18-27).

New independent Claim 63 contains the subject matter of new Claim 59 but includes the additional recitation of specific target proteins, as in Claim 62. New Claims 64-66, which are dependent on Claim 63, are phrased like new Claims 60, 61 and 62, and are similarly supported in the specification.

The amendments to the claims and the new claims do not add new matter. Applicants request their entry and favorable consideration.

The Objection to the Specification

The Examiner objected to the specification on page 3 of the Office Action because of the omission of SEQ ID NOs on page 22 and page 51 of the specification. The Applicants appreciate the Examiner's notice of this omission and have amended the specification to include SEQ ID NOs on these pages. The Applicants accordingly request withdrawal of this objection. In addition, the Applicants have amended the specification at page 10, lines 1 and 4, to recite "In one embodiment" to make clear that the target proteins recited are exemplary. With respect to the paragraph directed to protein C, the Applicants direct the Examiner's attention to the references cited in the accompanying information disclosure statement and to their belief that the claims, as amended, do not read on methods for identifying protein C muteins for the purpose of making mutein therapeutics whose therapeutic activity is mediated by cleaving a protein C substrate in the coagulation pathway.

The Applicants believe the amendments to the specification do not add new matter and therefore respectfully request their entry and favorable consideration.

The Rejections of the Claims

35 U.S.C. 112, first paragraph: Written Description

The Examiner has rejected Claims 1-16 and 45-58 on page 3, of the Office Action for lack of written description. Claims 8, 10 and 55 have been canceled. Thus, this rejection is moot in regard to these claims. Applicants request withdrawal of this rejection in view of the claim amendments and remarks below.

The Examiner cites *In re Ruschig*, 379 F.2d 990, 995 (CCPA, 1967) contending that the "laundry list disclosure of every possible moiety does not constitute a written description of every species in a genus because it would not 'reasonably lead' those skilled in the art to any particular species."

Applicants respectfully disagree. The instant claims are directed to methods for identifying mammalian mutein proteases. By definition, the activities of the new muteins to be identified are not described in advance, nor can they be. While Applicants have described in great detail the methods for generating and identifying mammalian mutein proteases, including the sites at which one might make mutations with some expectation that altered specificity or increased cleavage activity might result, the Applicants have also taught that random mutagenesis can be used to generate such muteins. Requiring the Applicants to recite where the muteins are mutated to result in a protease with increased cleavage activity and/or altered specificity for a substrate sequence would render the claimed method unnecessary.

The test for written description is whether the inventors had possession of the invention at the time of filing. Applicants submit that this test is met.

First, to speed allowance of claims drawn to important subject matter, all of the claims, as amended, recite either specific types of targets or specific targets that the identified mammalian mutein proteases will cleave or specific wild-type mammalian protease scaffolds from which the mutein proteases are derived, or both. Independent Claim 1 (and thus the claims that depend therefrom) specifically recites that the target protein to be cleaved is selected from the group consisting of a cell surface molecule that transmits an extracellular signal for cell proliferation, a cytokine, a cytokine receptor, and a signaling protein that regulates apoptosis. This group of target proteins is well known, with many characterized members, and there is no basis for the conclusion that choice of a desired protein target from within this group was not within the possession of the inventors. Claim 7 further limits Claim 1 by specifically reciting that the mutein protease is derived from a specified set of proteases; accordingly the Examiner cannot maintain that these specific proteases are not supported by the written description of the specification. Independent Claims 53 and 59 specifically recite that the mutein protease is derived from a specific, well-defined group of proteases (granzyme A, granzyme B, granzyme M, cathepsin, MTSP-1, elastase, chymase, tryptase, chymotrypsin, collagenase, Factor Xa, Protein C, plasma kallikrein, plasmin, trypsin, thrombin, complement factor serine proteases, papain,

ADAMTS13, endopeptidase, furin, cruzain and plasminogen activator). These specifically recited mammalian protease scaffolds are well known, well characterized proteins. As the specification makes clear, the specifically recited mammalian or human protease scaffolds are of known sequence. See specification p. 17, lines 28-29, and Table 2. Thus, there can be no doubt that the inventors had possession of the invention as it relates to these materials.

Further, all of the claims further recite that the target protein is a protein involved in a mammalian pathology. In addition, the structural determinants for the various specifically recited serine and cysteine proteases is known -- that is, because of the extensive characterization of serine and cysteine proteases, the inventors were in possession of detailed knowledge of the specific amino acid residues in the protease scaffold that determine substrate specificity, as is expressly set forth in Table 3. Possession of this detailed knowledge and its recitation in the specification does not, however, in any way translate into an obligation to place that information in the claims. The method of the invention allows one to identify a protease mutein that has the potential to provide therapeutic benefit because it cleaves a substrate in a target protein involved in a pathology. The claims have been amended to focus the method on target proteins or proteases or both that have never been targeted for therapeutic intervention by a protease or by the specific type of protease encompassed by the claims. Because the mutations required for the optimal mutein cannot be predicted with certainty in advance, the method of the invention is vital to the development of this new therapeutic area. Requiring the claims to recite the precise proteases used, the precise targets used, and the precise mutations in the protease, would render the claims meaningless and defeat the purpose, with respect to this important invention, of our patent system.

Moreover, the substrate sequence that the various protease scaffolds cleave was also in the inventors' possession -- as is also expressly set forth in Table 3 (right column).

Finally, with respect to the number of mutations made, the identity of the amino acid mutated, and the exact nature of the mutation, the Examiner has already, by restriction and election, focused the number of mutations to the range of 1-20, for which there is explicit written description support, and that the specification is replete with description about what residues in a protease can be mutated. Indeed, with respect to the latter point, the specification makes clear that, while in many instances mutations will be made in the S1 to S4 region, in other instances it is desirable to make mutations at other locations or even randomly throughout the sequence of

the protease (see, for example, pages 18 to 24 of the specification). Accordingly, there is written description support for this aspect of the invention as well. With respect to the identity of the mutated amino acid, the specification recites that the mutated amino acid is of course a different amino acid, and numerous illustrative examples of such mutations are described in the specification.

The amended and new claims can readily be distinguished from the claims at issue in *In re Ruschig*. The claims at issue in *Ruschig* were composition claims (not method claims such as those at issue here) drawn to benzenesulphonylureas of a general formula. The U.S. Court of Customs and Patent Appeals ruled that there was insufficient written description in Ruschig's application because, "[s]pecific claims to *single compounds* require reasonably specific supporting disclosure"[1] (Emphasis added). The *Ruschig* decision is not on point here -- in the instant case, the protease mutein scaffolds are expressly and specifically (i.e., not generically) recited in the specification, and each scaffold is of known sequence.

Further, *Ruschig* does not stand for the proposition that a listing of specifically contemplated moieties does not constitute a written description. On the contrary, such a listing was missing in the application at issue in *Ruschig* -- in that case the Court held that generic disclosure of a few specific compounds and a chemical formula with two R-groups that generically encompassed thousands of compounds was not sufficient specific written description for each of the thousands of compounds not specifically exemplified in the specification.

The instant claims are drawn to methods of identifying a protease mutein that cleaves a specific substrate sequence. Thus, by definition, those practicing the method do not know what mutein protease will best cleave the target sequence; the method provides an inventive means to provide such knowledge. Accordingly, the instant claims cannot be rejected because the specification does not teach what muteins should be used to cleave a particular target sequence. The Applicants have provided the means, the "blaze marks on the trees,"[2] for determining which residues may be most effective in mutating a protease to the desired specificity. For example, as the Examiner acknowledged on page 5 of the Office Action, and as stated on page 21, line 7 of the instant specification, residues 57, 102 and 195 by chymotrypsin numbering, are residues of interest in this regard. The specification teaches as well that there are other residues which can

[1] *In re Ruschig* at 994.
[2] *Id.* at 995.

influence specificity. The specification teaches that the specificity of serine proteases can be changed by mutation of residues 189, 190, 191, 192 and 226 for the substrate at site P1; residue 57 and residues in the loop between 58 and 64 and 99 for the substrate at site P2; residues 192, 217 and 218 for the substrate at site P3; and residues in the the loop between 168 and 180, 215 and 97-100 for the substrate at site P4. The specification teaches that the specificity of cysteine proteases can be changed by mutation of residues 25, 159, and 175 (see page 18, line 33) and residues 66, 67, 68, 133, 157, 160 and 205 (see page 19, line 7). In addition, to illustrate the method, various mutations in granzyme B at positions 99, 218, 192, 174 are described in the specification, and the resulting changes of substrate specificity described in Table 5 at pages 23-24 of the instant specification. Accordingly, the specification is replete with written description support for the proteases that can be mutated and the sites and nature of the mutations made in the methods claimed. Importantly, however, the method of the invention is not limited in any manner by where a protease is mutated, and it should not be. In the effort to develop important new therapeutic proteases that cleave target proteins not cleaved by them in their native environment, a wide variety of mutations may need to be examined, and the present method allows large libraries of protease muteins of any type to be screened for this important therapeutic activity.

Applicants also respectfully submit that the Examiner's comment, at page 5 of the Office Action, that "[t]he illustrative Examples, that provide the detailed description of the invention is drawn to a *single*, defined species of the component method" (emphasis added) is not well-founded.

First, the law is clear that the claims ought not to be limited to the working examples. The Court in *In re Rasmussen,* 650 F.2d 1212, 1215 (CCPA 1981) considered whether a written description disclosing a single example was insufficient to support broader claims and deemed as a matter of law that it was not. The court explained: "[T]hat a claim may be broader than the specific embodiment disclosed in a specification is in itself of no moment." *Id.* The court stated that "one skilled in the art who read Rasmussen's specification would understand that it is unimportant *how* the layers are adhered, so long as they are adhered."

Second, in the instant application, the specification demonstrates that the inventors were aware of numerous exemplary scaffolds, of known sequence, and had detailed knowledge of the specific amino acid residues in the protease scaffold that determine substrate specificity, as well as detailed knowledge of the substrate sequence that the various protease scaffolds cleave. Moreover, the specification teaches that such detailed knowledge is not required for the method of the invention to be applied to a particular protease and expressly recites that random mutagenesis can be employed to generate mutein proteases suitable for use in the method. Accordingly, the inventors were in possession of the invention as it applies to any protease and any target. Given that, to speed allowance of claims drawn to important subject matter, the claims are now limited to specific types of targets and/or specific proteases, or both, the Examiner is urged to reconsider and withdraw this rejection.

Accordingly, Applicants submit that the specification provides written description support for the claims as amended and new claims. This rejection should be withdrawn.

35 U.S.C. 112, first paragraph: Enablement

The Examiner has rejected claims 1-16 and 45-58 on page 6, of the Office Action for lack of written description. Claims 8, 10 and 55 have been canceled. Thus, this rejection is moot in regard to these claims. The Applicants respectfully request reconsideration and withdrawal of this rejection in view of the amendments to the claims and the remarks below.

The Examiner stated on page 7 of the Office Action that the specification failed to give adequate direction or guidance in how to determine the mutations that can be made to a scaffold of any protease to produce a library of muteins. Applicants disagree. The specification is replete with disclosure on the criteria for choosing a scaffold and engineering it to make protease muteins (see pages 16-24 of the specification). Requiring the claims to recite where to mutate a protease to obtain the protease to be identified by the method would render the method much less useful. The method is intended to help the artisan identify a protease mutein that has a new activity that makes it potentially useful as a therapeutic. If the artisan knew how to make such a mutein *de novo*, then the method would have much less value. Accordingly, while the specification provides guidance on the locations that might be mutated to alter activity or specificity, it also teaches that one can mutate the protease anywhere in the effort to create a library of muteins from which muteins with promising activity can be isolated. Based on this

specific guidance in the specification, and the general technical knowledge of making muteins, the ordinarily skilled artisan with the specification in hand could, without undue experimentation, make the recited mutein protease libraries and screen them in accordance with the method.

In addition, the specification details numerous specific protease scaffolds (see, e.g., Table 3) and these scaffolds are well characterized proteins, of known sequence with known cleavage domains and known substrate sequence recognition profiles -- so there can be no undue experimentation using those specifically recited protease scaffolds. Moreover, the specification elucidates well known mutagenesis techniques; again evidencing that no undue experimentation is required. The specification makes clear that one need not mutate a particular residue or residues of a protease but that random mutagenesis techniques can be employed (see, e.g., specification, p. 28, lines 17-23). One of the advantages of the instant method is that one need not know where to make a mutation to identify a protease mutein with therapeutic potential. Instead, one can simply make a library of muteins in which a variety of mutations are made and then screen them in accordance with the method to identify the protease mutein with the desired altered specificity and/or increased cleavage activity.

The Examiner also contends that the specification lacks working examples. The law does not require any working examples. Nonetheless, Applicants did provide a working example wherein numerous granzyme B mutants, and the substrate which they cleave, are described. The Examiner is referred to Table 5 on pages 23 and 24 of the specification. The instant specification also describes other sites for mutating serine and cysteine proteases, as described above. This is all that is required for enablement.

The Examiner stated on pages 7 and 8 of the Office Action that, because the breadth of the claims encompasses a large diversity of mutant proteins, and because it is well known in the art that it is difficult to know which mutations can be performed without deleteriously affecting protein function, the invention is not enabled. Applicants respectfully disagree. The claims recite a method of identifying a protease mutein for cleaving a target involved in a mammalian or human pathology. The fact that the ordinarily skilled artisan may test numerous muteins in the methods to identify useful candidates does not convert the routine testing clearly set forth in the specification into "undue experimentation". For these reasons, the Examiner's concerns do not support the contention that the method is not enabled.

The Examiner stated on pages 8 and 9 of the Office Action that Applicants' specification has not assured persons of skill in the art that the numerous variables of the claim would result in a mutation having pharmacologic activity without undue experimentation. The claims at issue do not require that the mutein identified be useful in treating a disease. Rather, the method claimed is a method for identifying a protease mutein derived from a mammalian or human protease or a specifically recited group of such protease scaffolds that can cleave a target protein at a desired substrate sequence, where the target protein is involved in a pathology. There is no recited claim limitation that the mutein derived have pharmacologic activity. The Applicants note for the Examiner's consideration that the language in Claim 1 "wherein cleavage of said substrate sequence in said target serves as a treatment for said pathology," is a limitation on the nature of the substrate sequence and not a requirement that the protease mutein have therapeutic activity *in vivo*. The specification teaches that the substrate sequence targeted by the protease must, when cleaved, alter the function of the target protease, and the language in the claims simply reflects that teaching and does not require that every protease mutein identified by the method have pharmacologic activity when administered to a patient. For example, the specification teaches at page 9, lines 17 - 21, "If a protease is engineered to recognize a substrate sequence within a target protein or proteins that would (*i.*) alter the function *i.e.* by inactivation of the target proteins(s) upon catalysis of peptide bond hydrolysis and, (*ii.*) the target proteins(s) are recognized or unrecognized as points of molecular intervention for a particular disease or diseases, then the engineered protease has a therapeutic effect via a proteolysis-mediated inactivation event." Accordingly, the claims simply require that the substrate sequence lie in the target protein at a site where cleavage of the target would result in an alteration of the activity of the target, such that, if such alteration occurred in vivo, a therapeutic benefit, relative to the pathology, would result. Numerous examples of such substrate sequences are provided in the application, see, e.g., pages 9 and 10 of the specification. This language distinguishes the claim from random alterations of specificity that might result in a protease that cleaves a target at locations having no effect on the activity of the target protein.

Applicants respectfully submit that the amended and new Claims are enabled and request that this rejection be withdrawn.

35 U.S.C. 112, second paragraph: Indefiniteness

The Examiner has rejected Claims 1-16 and 45-58 on page 10, of the Office Action for indefiniteness. Claims 8, 10, 49 and 55 have been canceled. Thus, this rejection is moot in regard to these claims.

The Examiner stated on page 10 of the Office Action that canceling the phrase "protease scaffold" rendered the claims indefinite. The Applicants have amended Claim 1, and the other independent claims have either been amended or presented with identical language, to clarify that the muteins are muteins of a mammalian protease and that the N mutations made are relative to "a wild-type scaffold sequence of said mammalian protease". This makes clear that the N mutations in the mutein protease sequence are relative to the wild-type protease scaffold (which may be a fragment of the wild-type mammalian protease).

The Examiner also stated on page 10 of the Office Action that "the activity" in part (b) of claim 1 lacks antecedent basis. The Applicants have amended Claim 1, and the other independent claims have either been amended or presented with identical language, to replace the phrase "the activity" with the phrase "an activity."

The indefiniteness rejections should be withdrawn.

35 U.S.C. 103 Rejection

The Examiner rejected claims 1-16 and 45-58 as obvious over either the Harris *et al.* reference or the Harris *et al.* reference in view of the Bianchi *et al.* reference. Claims 8 and 55 have been canceled. Thus, this rejection is moot in regard to these claims. The Examiner alleged that the Harris *et al.* reference discloses a method of identifying optimal substrate specificity for proteases, including granzyme B, using combinatorial methods of synthetic substrate libraries, and that the Harris *et al.* reference discloses two granzyme B variants, R192A and R192E (see Office action, paragraph bridging pages 11-12). The Examiner acknowledges that the Harris *et al.* reference does not disclose a combinatorial library mutant for the enzyme granzyme B (id.), but states that the Bianchi *et al.* reference discloses the use of peptide libraries in protease drug discovery and the advantages of a combinatorial library of enzymes. Applicants respectfully request that this rejection be reconsidered and withdrawn in view of the amendments to the claims and in view of the following remarks.

Independent Claims 1 and 53 have been amended, and all of the independent claims presented now require that the protease mutein identified cleaves a substrate sequence in a target

protein involved in a pathology, such that cleavage of the target protein at the substrate sequence would provide a treatment for the therapy. The Harris *et al.* reference relates to the identification of individual amino acids responsible for determining the stringent substrate specificity of granzyme B through the construction of a three-dimensional model of granzyme B complexed to substrate. This reference also describes two variants, each containing a single point mutation of arginine 192 (see the Harris *et al.* reference, page 27365) but does not suggest or disclose that the muteins made could have utility as agents for development in the treatment for a pathology by cleavage of a target protein involved in that pathology. Thus, Applicants assert that one of skill in the art would not be motivated by the teachings of Harris to generate and screen a library of mutein proteases as provided by the claims of the instant application. The Bianchi *et al.* reference does nothing to cure the deficiencies of the Harris *et al.* reference. The Bianchi *et al.* reference discloses viral enzyme conformational ensembles can be inhibited by a correspondingly large set of peptide inhibitors (see page 112, column 2). The Bianchi *et al.* reference does not teach or suggest a method for screening protease muteins to identify those that can cleave a substrate sequence in a target protein at a site that could confer a therapeutic benefit *in vivo*, as required by the pending claims, and nothing in either reference cited by the Examiner suggests that the two references should be combined for that purpose. Neither the viral enzyme conformational ensembles nor the peptide inhibitor libraries disclosed by the Bianchi *et al.* reference are analogous to the enzyme libraries recited in the current claims, and the Applicants cannot discern any basis for combining this reference with the Harris *et al.* reference or, even if combined, how the combination suggests the method of the instant claims.

All of the claims have been amended or newly presented to focus the method on either target proteins that have never been targeted for therapeutic intervention by a protease or on proteases that have never been used for therapeutic interevention at the target protein cleaved by them or both. The method is therefore drawn to a completely new field of drug discovery and provides an important new tool for the development of potentially life-saving therapeutic medicines. The method is, the Applicants respectfully submit, patentable under our laws.

For the above-stated reasons, the Applicants respectfully submit that the now pending claims would not have been obvious over the Harris *et al.* reference alone or in combination with the Bianchi *et al.* reference at the time the present invention was made. Accordingly, the Applicants respectfully submit that the rejection under 35 U.S.C. § 103 be withdrawn.

The Accompanying Information Disclosure Statement

Applicants file herewith an Information Disclosure Statement citing US Patent number 6,630,138 (the '138 patent). For the sake of completeness, Applicants address herein the distinctions between the currently claimed invention and the subject matter disclosed in the '138 patent.

The '138 patent teaches derivatives of the serine protease Protein C, which inactivates clotting factors Factor V_a and $VIII_a$ (see, the '138 patent, col. 1, lines 16-19). Applicants note that the amended claims and newly added claims either do not read on Protein C as a protease from which muteins screened in the method or derived or require that, if Protein C muteins are employed, they are being screened to identify a mutein that cleaves a target protein that is not a clotting factor. The '138 patent does not teach or disclose any protease muteins other than Protein C or in any way suggest that, as the present invention teaches, a protease can be mutated to increase its cleavage activity or alter its specificity such that the resulting mutein can be used for therapeutic intervention by cleaving a target that the wild-type protease from which it is derived is not known to cleave or cleaves with insufficient activity or specificity to enable its use as a therapeutic.

CONCLUSION

On the basis of the foregoing amendments and remarks, the Applicants respectfully submit that this paper is fully responsive and that the pending claims are in condition for allowance. Reconsideration of the application, withdrawal of all rejections, and allowance of now pending Claims 1-7, 9, 11-16, 45-48, 50-54, and 56-66 is respectfully requested.

The Commissioner is hereby authorized to charge payment of any fees required in connection with the papers transmitted herewith, or to credit any overpayment of same, to Deposit Account No. 50-0311, Reference No.25840-501. If there are any questions regarding these amendments and remarks, the Examiner is encouraged to contact the undersigned at the telephone number provided below.

Respectfully submitted,

Dated: September 27, 2005

Ivor R. Elrifi, Reg. No. 39,529
Naomi S. Biswas, Reg. No. 38,384
Attorneys for Applicants
c/o MINTZ, LEVIN, COHN, FERRIS, GLOVSKY AND POPEO, P.C.
Tel: (617) 542-6000
Fax: (617) 542-2241
Customer number 30623

TRA 2063542v5